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TOXIC SUBSTANCES

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DATE: **May 01, 2003**

MEMORANDUM

SUBJECT: **MANCOZEB**. 3rd Report of the Hazard Identification Assessment Review Committee.

FROM: Kit Farwell, D.V.M.
Reregistration Branch 1
Health Effects Division (7509C)

THROUGH: Jess Rowland, Co-Chair
and
Elizabeth Doyle, Co-Chair
Hazard Identification Assessment Review Committee
Health Effects Division (7509C)

TO: Tim Dole, C.I.H., Risk Assessor
Reregistration Branch 1
Health Effects Division (7509C)

PC Code: 014504

Mancozeb is an ethylenebisdithiocarbamate (EBDC) fungicide. Mancozeb was first evaluated by the HIARC on May 4, 1999 and was re-evaluated on September 18, 2001, to select endpoints consistent with new exposure durations. On February 20, 2003, the Health Effects Division (HED) Hazard Identification Assessment Review Committee (HIARC) reevaluated mancozeb in accordance with the 2002 OPP 10X guidance document. This report supercede the two previous HIARC reports on mancozeb.

Committee Members in Attendance

Members present were: Ayaad Assaad, William Burnam, Elizabeth Doyle, Pamela Hurley, John Liccione, Elizabeth Mendez, PV Shah, Jess Rowland, Brenda Tarplee, Bill Dykstra

Member(s) in absentia: Jonathan Chen, Paula Deschamp, Susan Makris

Data evaluation prepared by: Kit Farwell, D.V.M., Toxicologist

Also in attendance were: Bill Hazel, Chistina Swartz, Tim Dole (RRB1); and Anne Overstreet (SRRD)

Data Evaluation / Report Presentation

Kit Farwell

Kit Farwell, D.V.M.
Toxicologist

INTRODUCTION

Mancozeb is an ethylenebisdithiocarbamate (EBDC) fungicide. Mancozeb was first evaluated by the HIARC on May 4, 1999 and was re-evaluated on September 18, 2001, to select endpoints consistent with new exposure durations. On February 20, 2003, the Health Effects Division (HED) Hazard Identification Assessment Review Committee (HIARC) reevaluated mancozeb in accordance with the 2002 OPP 10X guidance document. This report supercede the two previous HIARC reports on mancozeb.

1. FQPA HAZARD CONSIDERATIONS

1. Adequacy of the Toxicity Data Base

There are acceptable developmental toxicity studies in rats and rabbits and an acceptable reproduction study in rats with mancozeb. There is a datagap for an acute neurotoxicity study with mancozeb and for a comparative study for thyroid toxicity in adults and offspring with mancozeb. A developmental neurotoxicity with ETU, a mancozeb metabolite, is also required.

2. Evidence of Neurotoxicity

Microscopic peripheral nerve injury at 50 mg/kg/day and above in a subchronic neuropathology study in rats included demyelination, myelin phagocytosis, Schwann cell proliferation, thickened myelin sheath, intrasheath ellipsoids, neurofibrillary degeneration, and myelin ovoids, bubbles, and debris. Affected nerves included the sciatic nerve in males and tibial and sural nerves in males and females. Related clinical signs at 339 mg/kg/day in males and 412 mg/kg/day in females included abnormal gait, limited use of rear legs and reluctance to walk, no use of rear legs, loss of posterior thigh muscle mass, and stained fur. Females in the 412 mg/kg/day group were discontinued from dosing after 2 weeks treatment because of high mortality; rats in this group recovered use of the rear legs after discontinuation of dosing.

Malformations of the central nervous system in a developmental toxicity study in rats at 512 mg/kg/day mancozeb included atrophy of brain tissue, cranial edema, dilated ventricles of the brain, compression and/or hemorrhages of the spinal cord, deficiency of tissue in the olfactory bulb, and meningoencephalocele. These malformations are believed to be due to the mancozeb metabolite, ETU, which caused the same malformations at lower doses.

3. Developmental Toxicity Study Conclusions

Rat Developmental Toxicity Study:

In a developmental toxicity study (MRID 00246663), **mancozeb** (83.0%) was administered to pregnant SD BR rats (26/dose group) by gavage at dose levels of 0, 2, 8, 32, 128, or 512 mg/kg/day from days 6-15 of gestation. A separate group received 50 mg/kg/day of **ethylene thiourea** (ETU, 99%), a mancozeb metabolite and degradate. Doses for mancozeb and ETU were calculated on day 6 and not adjusted for weight changes during the study. No data on the stability of ETU were reported.

Maternal toxicity in the 512 mg/kg/day **mancozeb** group included one maternal death attributed to treatment and 2 dams sacrificed due to abortion. Maternal clinical signs in the 512 mg/kg/day mancozeb group included lethargy, scruffy coat, and diarrhea. Food consumption was decreased significantly among dams in the 128 mg/kg/day (-11%) and the 512 mg/kg/day (-88%) mancozeb groups during days 10-15 of gestation when compared to controls. Food consumption remained depressed somewhat during the post-treatment period. Body weights were decreased in the 128 mg/kg/day (-11%) and the 512 mg/kg/day (-25%) mancozeb groups when compared to controls on day 20 of gestation. Body weight gains were decreased compared to controls at 128 mg/kg/day (-51%) and weight loss occurred at 512 mg/kg/day during several weekly intervals. Pregnancy rates were comparable among groups.

Maternal survival and body weights in the 50 mg/kg/day **ETU** group were comparable to controls while food consumption was increased in comparison to controls. Body weight gains were decreased in the ETU group (-36%) compared to controls during the treatment period.

Developmental toxicity in the 512 mg/kg/day **mancozeb** group included increased mean resorptions (3.77/dam compared to 0.39/dam in controls) and an increased number of dams with total resorptions (9/22 vs 0/23 pregnant dams in controls) which resulted in a decreased number of live litters (13 vs 23 in controls). Mean implants were decreased slightly in the 512 mg/kg/day mancozeb group (10.82 vs 12.48 in controls). Mean numbers of live fetuses per dam were similar between groups. The 512 mg/kg/day group had decreased fetal weight (-18% compared to controls) and an accompanying decrease in gravid uterine weight. The number of implantation sites in the control group was erroneously reported as greater than the number of corpora lutea. Developmental abnormalities attributed to treatment with 512 mg/kg/day mancozeb included abnormalities of the **central nervous system:** atrophy of brain tissue, cranial edema, dilated ventricles of the brain, compression and/or hemorrhages of the spinal cord, deficiency of tissue in the olfactory bulb, meningoencephalocele; abnormalities of the **skeletal system:** incomplete cranial ossification, wide cranial sutures, curved clavicle, fused sternebrae, absent caudal or sacral vertebrae, fused and/or thickened ribs, wavy ribs, misshapen or incomplete ossification of hindlimb long bones, kyphosis, incomplete ossification or misshapen pelvis; **gross defects:** agnathia, cleft palate, cleft lip, club limb, stubby tail, forelimb flexure, and kinked tail; and cryptorchidism.

The **ETU** group (50 mg/kg/day) had decreased mean fetal body weight (-13% in comparison to controls). The number of implants, resorptions, and litter size were all similar to control values. Developmental abnormalities attributed to treatment in the ETU group included the above named central nervous system defects, skeletal defects, gross defects, and cryptorchidism noted with mancozeb, as well as exencephaly, ectopic kidneys, agenesis of the kidneys, hydronephrosis, reduced stomach with thickened wall, edematous fat pads, incomplete ossification of ribs, less than 13 ribs, fused lumbar, sacral, or caudal vertebrae, abnormal pelvic limb posture, oligodactyl, syndactyl, webbed digits, and anal atresia.

Although mancozeb and ETU caused many of the same developmental defects, with the exception of total resorptions, ETU was in general a more severe developmental toxicant than mancozeb because: (a) a smaller dose of ETU (50 mg/kg/day) caused developmental defects than did mancozeb (512 mg/kg/day); (b) many of the same developmental defects occurred with greater frequency with ETU than with mancozeb; (c) more types of developmental defects occurred with ETU than with mancozeb; and (d) developmental defects which occurred with ETU were accompanied by minimal maternal toxicity in comparison to mancozeb.

The **maternal NOAEL for mancozeb** is 32 mg/kg/day and the **maternal LOAEL** is 128 mg/kg/day based on decreased food consumption, body weight, and body weight gains.

The **developmental NOAEL for mancozeb** is 128 mg/kg/day and the **developmental LOAEL** is 512 mg/kg/day based on gross developmental defects, central nervous system defects, skeletal defects, cryptorchidism, abortions, increased resorptions, and decreased fetal weight.

Maternal and developmental NOAELs for **ETU** were not determined because only 1 dose of ETU was used. The maternal LOAEL for ETU was 50 mg/kg/day based on decreased body weight gain. The developmental LOAEL for ETU was 50 mg/kg/day. Developmental toxicity at that dose included gross developmental defects, central nervous system defects, skeletal defects, cryptorchidism, and decreased fetal weight.

This study is **acceptable/guideline** for a developmental toxicity study in rats and **satisfies** the guideline requirement for a developmental toxicity study with **mancozeb**.

Rabbit Developmental Toxicity Study:

In a developmental toxicity study (MRID 40433001), mancozeb (83.0%) was administered to pregnant NZW rabbits (20/dose group) by gavage at dose levels of 0, 10, 30, or 80 mg/kg/day from days 7 through 19 of gestation.

Treatment-related maternal mortality occurred to two does in the 80 mg/kg/day group. Clinical signs of maternal toxicity in high-dose does during the treatment period included alopecia, anorexia, scant feces, anuria, ataxia, reddish discharge on cage liner, and stained perineum. Most affected does showed similar toxicity during the post-treatment period (days 20-29). Five does in the 80 mg/kg/day group aborted and had decreased food consumption and body weight losses. With the exception of these 5 does, comparable body-weight changes were observed among the groups during and after treatment.

There were no treatment-related increases in type or incidence of developmental variations or fetal malformations. Mean fetal body weights were comparable among the groups. There were no treatment-related effects on pregnancy rate, litter size, number of resorptions, mean number of corpora lutea, implantations, fetal deaths, or sex ratios.
The **maternal NOAEL** is 30 mg/kg/day. Toxicity at the **maternal LOAEL** of 80 mg/kg/day included abortions, mortality, and clinical signs.

The **developmental NOAEL** is 30 mg/kg/day and the **developmental LOAEL** is 80 mg/kg/day based on abortions.

This study is classified **acceptable/guideline** and **satisfies** the guideline requirement for a developmental toxicity study in rabbits.

4. Reproductive Toxicity Study Conclusions

In a 2-generation reproduction study (MRID 41365201), mancozeb (84%) was administered to Crl:CD BR rats (25/sex/dose group) in diet at dietary dose levels of 0, 30, 120, or 1200 ppm. P_1 rats initially received dietary concentrations that were ½ of these dietary concentrations before adjustment to the final concentration after 4 weeks of treatment. Doses were equivalent to 0, 1.73, 6.95, 68.90 mg/kg/day for males and 0, 1.83, 7.47, or 79.37 mg/kg/day for females. Two litters were weaned by both the P_1 and P_2 parental generations.

There were no treatment-related effects on survival and clinical signs were comparable among groups in both generations. Mean body weights were decreased in comparison to controls in 1200 ppm males (-12% for P_1 and -7% for P_2 generations) and females (-16% for P_1 and -14% for P_2 generations) at termination. Food consumption was decreased in P_1 males (-7%) and P_1 females (-10%) but was not changed in the P_2 generation.

Increased relative thyroid weights in 1200 ppm males and females of both generations were accompanied by an increased incidence and/or severity of follicular cell hyperplasia. Thyroid follicular cell adenomas were found in 1200 ppm males in both the P_1 generation (3/25 vs 0/25 in controls) and P_2 generation (4/25 vs 0/25 in controls), but without statistical significance.

Absolute and relative liver weights were decreased in 120 ppm parental animals, except for P_1 males which had increased absolute liver weight (+5%, not statistically significant) and increased relative liver weight (+6%, statistically significant). Relative liver weights in 1200 ppm males and females of both generations were increased (+12 to 16%) while absolute liver weights were variably increased or decreased. Increased relative liver weights in 1200 ppm animals is attributed to treatment, but is considered an equivocal effect in 120 ppm P_1 males in light of the decreased liver weights in other 120 ppm groups. Prominent lobular liver architecture was seen grossly in 1200 ppm males and females, but no histopathological liver lesions occurred.

Brown globular pigment was found in proximal tubular lumens of kidneys of both sexes and generations in 120 and 1200 ppm groups. This pigment (negative for iron) was limited to the luminal space, did not damage tubular epithelium and is not considered toxicologically significant in the absence of pathological changes.

The parental NOAEL/LOAEL are increased from 30/120 ppm in the previous review because increased relative liver weight in 120 ppm P_2 males is considered an equivocal effect and the pigment in renal tubular lumen is not considered toxicologically significant. The **parental NOAEL** is now 120 ppm (males: 6.95 mg/kg/day; females; 7.47 mg/kg/day) and the **parental LOAEL** is now 1200 ppm (males: 68.90; females: 79.37 mg/kg/day) based upon body weight decrements, increased relative thyroid weights, and increased incidence of thyroid follicular cell hyperplasia.

No adverse reproductive or offspring effects were attributed to mancozeb. Fecundity and gestation indices; litter sizes; and pup viability, survival, and body weights were all similar among the groups. The **NOAEL** for **reproductive** effects is $\geq$ 1200 ppm (males: 69.90 mg/kg/day; females: 79.37 mg/kg/day), the highest dose tested.

This study is classified **acceptable/guideline** and **satisfies** the guideline requirement for a reproduction study in rats.

5. Additional Information from Literature Sources

Manganese and carbon disulfide are risk factors for developmental of Parkinson's disease in humans. Since mancozeb and maneb contain manganese, and also degrade to carbon disulfide, there has been speculation in the literature as to whether exposure to EBDC pesticides may play a role in the development of Parkinson's disease. Epidemiological studies have not shown a relationship to Parkinson's from use of EBDC pesticides, though pesticide use in general has been shown to be a risk factor (Tanner, C; Gorell, J; Checkoway, H. Epidemiological studies: Risk factors session. Parkinson's Disease, Environment and Genes. Nineteenth International Neurotoxicology Conference. 2001. NeuroToxicology 22: 837-844).

6. Pre-and/or Postnatal Toxicity

The HIARC concluded that there is not a concern for pre- and/or postnatal toxicity resulting from exposure to mancozeb in the reproduction study in rats or in the developmental toxicity studies in rats and rabbits.

A. Determination of Susceptibility

There was no evidence of increased quantitative susceptibility of fetuses in the rat developmental study because the maternal NOAEL was lower than the developmental NOAEL: the maternal NOAEL was 32 mg/kg/day and the maternal LOAEL was 128 based on decreased food consumption, body weight, and body weight gains; the developmental NOAEL was 128 mg/kg/day and the developmental LOAEL was 512 mg/kg/day based on gross developmental defects, central nervous system defects, skeletal defects, cryptorchidism, abortions, increased resorptions, and decreased fetal weight. There was no evidence of increased qualitative susceptibility in the rat developmental study because malformations were accompanied by severe maternal toxicity (one maternal death, presence of clinical signs, and mean body weight decrements of -25% compared to controls).

There was no indication of increased susceptibility (quantitative or qualitative) of fetuses in the rabbit developmental study: the developmental NOAEL was 30 mg/kg/day based on abortions; maternal toxicity at the same dose included mortality and clinical signs.

There was no indication of increased susceptibility (quantitative or qualitative) to offspring in the 2-generation reproduction study in rats: the parental NOAEL was 7 mg/kg/day and the parental LOAEL was 69 mg/kg/day based upon body weight decrements, increased relative thyroid weights, and increased incidence of thyroid follicular cell hyperplasia; the reproductive NOAEL was $\geq$ 69 mg/kg/day, the highest dose tested.

B. Degree of Concern Analysis and Residual Uncertainties

There are no concerns nor residual uncertainties for pre- and/or postnatal toxicity from exposure to mancozeb.

C. Special FQPA Safety Factor(s):

Based upon the above-described data, no special FQPA Safety Factor is needed (i.e. 1X) since there are no residual uncertainties for pre and/or post natal toxicity.

The Special FQPA Safety Factor recommended by the HIARC assumes that the exposure databases (dietary food, drinking water, and residential) are complete and that the risk assessment for each potential exposure scenario includes all metabolites and/or degradates of concern and does not underestimate the potential risk for infants and children.

7. Recommendation for a Developmental Neurotoxicity Study

The HIARC concluded that there is a concern for developmental neurotoxicity resulting from exposure to mancozeb because of teratogenic effects to the brain in the developmental toxicity studies in rats with mancozeb and with the mancozeb metabolite, ETU. Although the HIARC determined previously that a developmental neurotoxicity (DNT) study was required for mancozeb (HED Doc. 0050362), this requirement will now be addressed by a DNT study with the mancozeb metabolite, ETU, because the teratogenic effects to the brain after mancozeb treatment are believed to be due to ETU.

Mancozeb affects thyroid hormone homeostasis following oral and dermal exposure (increased thyroid weight, decreased T4, and follicular cell hyperplasia) in subchronic and chronic studies. Additionally, the mancozeb metabolite, ETU, has been shown to be a direct-acting thyroid toxicant by inhibiting thyroid peroxidase enzyme. The dose response for thyroid toxicity is well characterized in a number of studies with adult animals, however, there are no data for thyroid hormones in the young and no information as to whether the young are more sensitive than adults with respect to this endpoint. Since the doses and endpoints selected for overall risk assessments are based on thyroid toxicity, there are no residual uncertainties with regard to thyroid toxicity, however, the HIARC concluded that a comparative assessment of the thyroid using young and adult animals is required to address the concern for thyroid effects on the young animal. The Task Force should consult the Agency on the protocol for this study before beginning the study.

The HIARC determined that a 10X database uncertainty factor (UF_{DB}) is needed to account for the lack of these studies since the available data provide no basis to support reduction or removal of the default 10X factor.

II. HAZARD IDENTIFICATION

1. Acute Reference Dose (aRfD): Females 13+ years of age

Study: Developmental Toxicity in Rabbits
OPPTS 870.3700 (OPP §83-3b)

MRID No.: 40433001

Executive Summary:

In a developmental toxicity study (MRID 40433001), mancozeb (83.0%) was administered to pregnant NZW rabbits (20/dose group) by gavage at dose levels of 0, 10, 30, or 80 mg/kg/day from days 7 through 19 of gestation.

Treatment-related maternal mortality occurred to two does in the 80 mg/kg/day group. A death in the mid-dose group was due to dosing error and was not treatment related. Clinical signs of maternal toxicity in high-dose does during the treatment period included alopecia, anorexia, scant feces, anuria, ataxia, reddish discharge on cage liner, and stained perineum. Most affected does showed similar toxicity during the post-treatment period (days 20-29). Five does in the 80 mg/kg/day group aborted and had decreased food consumption and body weight losses. With the exception of these 5 does, comparable body-weight changes were observed among the groups during and after treatment.

There were no treatment-related increases in type or incidence of developmental variations or fetal malformations. Mean fetal body weights were comparable among the groups. There were no treatment-related effects on pregnancy rate, litter size, number of resorptions, mean number of corpora lutea, implantations, fetal deaths, or sex ratios.

The maternal NOAEL is 30 mg/kg/day. Toxicity at the maternal LOAEL of 80 mg/kg/day included abortions, mortality, and clinical signs. The developmental NOAEL is 30 mg/kg/day and the developmental LOAEL is 80 mg/kg/day based on abortions. This study is classified acceptable/guideline and satisfies the guideline requirement for a developmental toxicity study in rabbits.

Dose and Endpoint for Establishing aRfD: NOAEL = 30 mg/kg/day based on abortions at 80 mg/kg/day (LOAEL).

Uncertainty Factor (UF): 1000 (10 for interspecies extrapolation, 10 for intraspecies variations, and 10 for a database uncertainty factor for the lack of a comparative thyroid study with mancozeb and a developmental neurotoxicity study with the mancozeb metabolite, ETU).

Comments about Study/Endpoint/Uncertainty Factor: Abortions are an effect which may occur after exposure of 1 day duration and are thus appropriate for this population/subgroup. Abortions also occurred in the rat developmental study at a higher dose.

$$\textbf{Acute RfD (Females 13+)} = \frac{30\text{ mg/kg (NOAEL)}}{1000\text{ (UF)}} = \textbf{0.03 mg/kg/day}$$

2. Acute Reference Dose (aRfD): General Population, including Infants and Children

Study Selected: None

MRID No.: N/A

Executive Summary: N/A

Dose and Endpoint for Establishing aRfD: N/A

Comments about Endpoint: An endpoint attributable to a single exposure was not available from oral studies, including the developmental toxicity studies.

3. Chronic Reference Dose (cRfD)

Study: Combined Chronic Toxicity/Carcinogenicity Rat Feeding Study
OPPTS 870.4300 (OPP §83-5)

MRID No.: 41903601

Executive Summary:

In a combined chronic toxicity/carcinogenicity study (MRID 41903601), mancozeb (83.8%) was administered to Crl:CD(BR) rats (72/sex/dose) for 2 years. Initial dietary concentrations of 0, 10, 30, 65, or 375 ppm were gradually increased to the final dietary concentrations of 0, 20, 60, 125, or 750 ppm (males: 0, 0.77, 2.33, 4.38, or 30.9 mg/kg/day; females: 0, 1.06, 3.06, 6.72, or 40.2 mg/kg/day) for weeks 5-104 so that young rats would receive similar doses to mature rats. There was an interim sacrifice of 10 rats/sex/dose at 1 year with the remaining 62 rats/sex/dose sacrificed after 2 years of treatment.

There were no treatment-related effects upon mortality, clinical signs, hematology, urinalysis, or clinical chemistry, other than thyroid hormones as detailed below. Decreased body weight occurred in high-dose males (-12%) and females (-10%) at various time points compared to controls. Body weight gains for weeks 1-91 were slightly decreased in high-dose males (+256% vs 276% in controls) and females (+124% vs 145% in controls).

Relative liver weight was significantly increased in high-dose males (+29%) and absolute liver weight was increased non-significantly (+11%). Relative testes weight was increased in high-dose males at 12 months (+17%). An increased incidence and severity of bilateral retinopathy occurred in 125 ppm females and 750 ppm males and females. The incidence of unilateral retinopathy was decreased in 125 and 750 ppm females.

Thyroid toxicity in high-dose males and females included changes in thyroid hormone levels, microscopic changes, and changes in thyroid weights. Thyroxine levels were decreased beginning at 3 months and remained decreased until 18 months in males and to study termination in females. Triiodothyronine was decreased at 3 months in both sexes but rebounded at 3 months and remained elevated throughout the study, sometimes with statistical significance. Thyroid stimulating hormone was elevated beginning at 6 months until study termination, sometimes with statistical significance. Absolute and relative thyroid/parathyroid weights were increased at 24 months but not at 12 months. Microscopically, the incidence of thyroid follicular cell hypertrophy/hyperplasia was increased at 2 years, but not at 1 year, in 750 ppm males and females. The incidence of thyroid nodular follicular cell hyperplasia was also increased in 750 ppm males and females at 2 years. Thyroid follicular cell adenomas were increased in high-dose males (0/60, 1/62, 1/60, 0/58, and 20/61) and females (1/62, 1/60, 1/62, 1/61, and 6/61). Thyroid follicular cell carcinomas were increased in high-dose males (0/60, 0/62, 2/60, 2/58, 14/61) and females (0/62, 0/60, 0/62, 1/61, 4/61).

A granular yellowish-brown pigment was seen microscopically in the kidneys of male and female rats receiving 125 or 750 ppm. The pigment was unaccompanied by histopathological lesions or clinical chemistry or urinalysis abnormalities. The NOAEL/LOAEL were previously based upon this pigment, however, the pigment is not considered toxicologically significant in the absence of pathological changes and the NOAEL/LOAEL are increased from 60/125 ppm in the previous review to 125/750 ppm.

The NOAEL is now 125 ppm (male: 4.38 mg/kg/day, female: 6.72 mg/kg/day) and the LOAEL is 750 ppm (male: 30.9 mg/kg/day, female: 40.2 mg/kg/day) based upon thyroid toxicity and bilateral retinopathy.

Based upon toxicity, doses were adequate to assess carcinogenicity. Although survival in the 5 male dose groups was low (26%, 29%, 39%, 16%, and 21%), there were enough survivors to assess carcinogenicity. This study is classified acceptable/guideline and satisfies the guideline requirement for a combined chronic toxicity/carcinogenicity study in rats.

<u>Dose Selected for Establishing Chronic RfD:</u> NOAEL = 4.38 mg/kg/day based on thyroid toxicity (microscopic changes, increased thyroid/parathyroid weight, and alterations in thyroid hormones) at the LOAEL of 30.9 mg/kg/day.

$$\textbf{Chronic RfD} = \frac{4.38\ \text{mg/kg/day}\ \ (\text{NOAEL})}{1000\ \ \ (\text{UF})} = \textbf{0.004mg/kg/day}$$

Uncertainty Factor (UF): 1000 (10 for interspecies extrapolation, 10 for intraspecies variations, and 10 for a database uncertainty factor for the lack of a comparative thyroid study with mancozeb and a developmental neurotoxicity study with the mancozeb metabolite, ETU)

Comments about Study/Endpoint: The duration of exposure in this study is appropriate for a chronic dietary risk assessment. The NOAEL in this study is based upon thyroid toxicity which is a consistent finding in other studies with mancozeb.

4. Incidental Oral Exposure: Short-Term (1-30 days)

Study: Subchronic rat study (13 weeks)
OPPTS 870.3100 (OPP §82-1a)

MRID No.: 00261536

Executive Summary:

In two subchronic toxicity studies (1986, MRID 00261536), mancozeb (84%) or ethylene thiourea (99.8%) was administered to Crl-CD(SD) rats (14/sex/group) in the diet for 13 weeks. Dietary concentrations of **mancozeb** were 0, 30, 60, 125, 250, or 1000 ppm (males: 1.78, 3.49, 7.42, 14.98, 57.34 mg/kg/day; females: 0, 2.20, 4.38, 9.24, 17.82, 74.64 mg/kg/day). A single dose group received **ethylene thiourea (ETU)**, a mancozeb metabolite, in the diet at a concentration of 250 ppm (males: 14.28 mg/kg/day; females: 17.81 mg/kg/day). Dietary concentrations were gradually increased throughout the study to maintain approximately the same compound intake in mg/kg/day, with dietary concentrations kept at the above levels for the last 8 weeks of the study.

Body weights in the 1000 ppm mancozeb and the 250 ppm ETU groups had decreases of similar magnitude when compared to controls, at most time points. Maximal body weight decrements in the 1000 ppm mancozeb group were -8% for males and -14% for females and in the 250 ppm ETU group were -6% for males and -8% for females. Body weight gain at termination in the 1000 ppm mancozeb group was decreased -12% in males and -13% in females in comparison to controls. Body weight gain at termination in the 250 ppm ETU group was decreased -6% in males and -12% in females in comparison to controls. Food consumption was significantly decreased at most time points for males in both the 1000 ppm mancozeb and 250 ppm ETU groups, but was only occasionally decreased for females in the same dose groups.

Thyroid toxicity, including changes in hormone levels, histopathology, and increased thyroid weights, occurred in males and females, principally in the 1000 ppm mancozeb and the 250 ppm ETU groups. Thyroxine levels were decreased in the 250 ppm mancozeb group (females: -28%), in the 1000 ppm mancozeb group (males: -34%; females: -43%), and in the 250 ppm ETU group (males: -50%; females: -65%) when compared to controls. Triiodothyronine levels were unchanged with mancozeb treatment but were increased in the 250 ppm ETU group (males: +28%; females: +16%) in comparison to controls. Thyroid stimulating hormone was increased in the 1000 ppm mancozeb group (males: +261%; females: +170%) and the 250 ppm ETU group (males: +408%; females: +263%) in comparison to controls. Absolute thyroid weights were increased in males in the 1000 ppm mancozeb group (+32%) and in the 250 ppm ETU group (males: +80%; females: +68%). Relative thyroid weights were increased in the 1000 ppm mancozeb group (males: +49%; females: +33%) and the 250 ppm ETU group (males: +84%; females: +80%). Thyroid follicular cell hyperplasia was seen microscopically in males and females in the 1000 ppm mancozeb and the 250 ppm ETU groups.

Increased relative liver weights occurred in the 1000 ppm mancozeb (males: +11%; females: +24%) and the 250 ppm ETU group (males: +12%; females: +15%). Absolute liver weights, in comparison to controls, were variable in the 1000 ppm mancozeb group (males: +2%, females: +16%) and the 250 ppm ETU group (males: +10%; females: +7%). Three males in the 250 ppm ETU group had grossly enlarged livers. Microscopically, there was an increased incidence of centrilobular hepatocyte hypertrophy in males in the 1000 ppm mancozeb group and males and females in the 250 ppm ETU group. Hepatic MFO aminopyrine N-demethylation enzyme activity was significantly reduced (-32%) in males in the 250 ppm ETU group. Aniline hydroxylation MFO activity was non-significantly reduced in males (-31%) and females (-40%) in the 1000 ppm mancozeb group.

Serum cholesterol was increased in males and females in the 250 ppm ETU group. Yellow-brown pigment in renal tubular cells was increased in 60 ppm and greater mancozeb groups but not in the ETU group. The pigment was attributed to the ethylenebisisothiocyanate metabolite of mancozeb and was unaccompanied by pathological changes.

Urine, blood, and thyroids were analyzed for ethylenebisdithiocarbamate and ETU. The majority of mancozeb was metabolized to ETU and excreted in the urine. ETU residues, but no EBDC residues, were detected in the thyroid.

The NOAEL/LOAEL for mancozeb were previously considered to be 60/125 ppm based upon renal pigment which is not considered toxicologically significant. The **NOAEL** for **mancozeb in females** is now 125 ppm (9.24 mg/kg/day) and the **LOAEL** is 250 ppm (17.82 mg/kg/day) based upon decreased serum thyroxine levels. The **NOAEL** for **mancozeb in males** is 250 ppm (14.98 mg/kg/day) and the **LOAEL** is 1000 ppm based upon body weight decrements, changes in thyroid hormones, changes in liver enzymes, microscopic changes in the liver and thyroids, increased absolute and relative thyroid weights, and increased relative liver weights.

A NOAEL for **ETU** was not established since only a single dose group was used; the LOAEL for ETU was 14.28 mg/kg/day.

This study is classified **acceptable/guideline** and **satisfies** the requirement for a subchronic study in rats with mancozeb.

<u>Dose and Endpoint for Risk Assessment:</u> NOAEL = 9.24 mg/kg/day based upon decreased serum thyroxine levels in females.

Comments about Study/Endpoint: This endpoint is appropriate for the population (infants and children) and exposure duration (1-30 days). Although the endpoint is based upon decreased thyroxine level at 13 weeks, the Committee believes that the decrease could occur within 30 days. This endpoint is supported by fact that thyroid lesions were seen after dermal exposure for 21 days with maneb and the thyroid is the target organ in a variety of species and studies. The NOAEL from the subchronic dog study (2.98 mg/kg/day) was not used because signs of toxicity at this dose were vague and not considered sufficiently robust for risk assessment.

5. Incidental Oral Exposure: Intermediate-Term (1 - 6 Months)

Study: Subchronic rat study (13 weeks)
OPPTS 870.3100 (OPP §82-1a)

MRID No.: 00261536

Executive Summary: See short-term incidental oral exposure.

Dose and Endpoint for Risk Assessment: NOAEL = 9.24 mg/kg/day based upon decreased serum thyroxine levels in females.

Comments about Study/Endpoint: This endpoint is appropriate for the population (infants and children) for the exposure duration (1-6 months). For additional comments, see the short-term incidental oral section of this document.

6. Dermal Absorption

Dermal Absorption Factor: 1% (from a dermal absorption study in rats, MRID 45802101)

Dermal Absorption Factor: A guideline dermal absorption study with mancozeb suggested a dermal absorption factor of 1%. Although the guideline dermal absorption study (MRID 45802101) reported < 1% dermal absorption after 8 hours, a value of 1% was used because this is an upper bound estimate and the methodology used is no more precise than ± 1% accuracy. The results of two non-guideline studies supported the value of 1% dermal absorption (MRID # 40955401 and Accession # 250063). The dermal absorption value is also supported by comparison of NOAEL values between the 28-day dermal toxicity study in rats and the 13-week rat feeding study (1000 mg/kg/day ÷ 9.25 mg/kg/day = 0.9% dermal absorption). The dermal absorption factor of 1% was also consistent with dermal absorption factors for the other EBDCs.

Executive Summary:

In a dermal penetration study (MRID 45802101) Mancozeb (86.4% a.i. batch #9903-261/311) was administered to four male Sprague-Dawley (Crl:CDBR) rats/dose/exposure duration to a 12 cm^2 shaved area of the back at dose levels of 12 and 0.014 mg/cm^2 Mancozeb in the 80WP formulation in water. Exposure duration was 8 hours and animals were monitored for 8, 72 and

144 hours. 72 and 144 hour groups were washed at 8 hours. Mean recoveries of mancozeb ranged from 93-99%. The mean percent dermal absorption of mancozeb at 8 hours was 0.05% after treatment at 8.00 mg/cm^2(~40 mg/kg) and was 0.12% after treatment at 0.013 mg/cm (~0.06mg/kg). Thyroids were collected from all animals and analyzed. Radioactivity was below the limit of detection (did not exceed background) for both doses.

This study in the rat is **acceptable** and satisfies the guideline requirement for a dermal penetration study (870.7600) in rats.

7. Dermal Exposure: Short-Term (1- 30 days) Exposure: Females 13+ years of age

Study: Subchronic rat study (13 weeks)
OPPTS 870.3100 (OPP §82-1a)

MRID No.: 00261536

Executive Summary: See short-term incidental oral exposure.

Dose and Endpoint for Risk Assessment: NOAEL = 9.24 mg/kg/day based upon decreased serum thyroxine levels in females.

Comments about Study/Endpoint: The 28-day dermal study with mancozeb was not used because thyroids were not examined microscopically and thyroid histopathology was seen after 21 days of treatment with maneb. Although the endpoint is based upon decreased thyroxine level at 13 weeks, the Committee believes that the decrease could occur within 30 days. This endpoint is supported by the fact that thyroid lesions were seen after dermal exposure for 21 days with maneb and the thyroid is the target organ in a variety of species and studies. The NOAEL from the subchronic dog study was not used because signs of toxicity at this dose were vague and not considered sufficiently robust for risk assessment.

8. Dermal Exposure: Intermediate-Term (1 - 6 Months): Females 13+ years of age

Study: Subchronic rat study (13 weeks)
OPPTS 870.3100 (OPP §82-1a)

MRID No.: 00261536

Executive Summary: See short-term incidental oral exposure.

Dose and Endpoint for Risk Assessment: NOAEL = 9.24 mg/kg/day based upon decreased serum thyroxine levels in females.

Comments about Study/Endpoint: the NOAEL is based upon toxicity to the thyroid which was a common effect seen in a variety of studies and species. The 28-day dermal study with mancozeb was not used because thyroids were not examined microscopically and thyroid histopathology was seen after 21 days of treatment with maneb. Furthermore, thyroid toxicity was not seen after 28 days of dermal exposure, but was seen after 90 days of exposure in the oral study. The NOAEL from the subchronic dog study was not used because signs of toxicity at this dose were vague and not considered sufficiently robust for risk assessment.

9. Dermal Exposure Long-Term (> 6 Months)

Study: Combined Chronic Toxicity/Carcinogenicity Rat Feeding Study
OPPTS 870.4300 (OPP §83-5)

MRID No.: 41903601

Executive Summary: See chronic dietary section of this document.

Dose and Endpoint for Risk Assessment: NOAEL = 4.38 mg/kg/day based on thyroid toxicity (microscopic changes, increased thyroid/parathyroid weight, and alterations in thyroid hormones) at the LOAEL of 30.9 mg/kg/day.

Comments about Study/Endpoint: This dose/endpoint/study was also selected to establish the chronic RfD.

10. Inhalation Exposure: Short -Term (1- 30 days):

Study: 90-Day Inhalation Study in Rats
OPPTS 870.3465 (OPP § 82-4)

MRID No.: Accession 00159471

Executive Summary:

In a subchronic inhalation toxicity study (MRID 00159471, final report and MRID 00261539, interim report), mancozeb (83.35%) was administered to groups of Crl:CD(SD)BR rats (38/sex/dose group) by nose-only exposure for 6 hours/day, 5 days/week for 13 weeks. Analytical concentrations were 0, 0.018, 0.079, or 0.326 mg/L. There was an interim sacrifice of 5/sex/group after 4 weeks, a sacrifice of 16/sex/group when dosing was terminated at 13 weeks, and a recovery group of 17/sex/group were held for an extra 13 weeks after the termination of dosing. Mass median aerodynamic diameters (MMAD) were ≤ 4.4 microns and geometric standard deviations (GSD) were ≤ 2.3.

The main study was preceded by a **rangefinding** study in which groups of Crl:CD(SD)BR rats (12/sex/dose-group) were exposed to mancozeb for 6 hours/day for 10 days by either whole-body or nose-only exposure. Analytical concentrations were 0.023, 0.138, and 0.519 mg/L; MMADs were 3.5-4.9 µm and GSDs were 2.2-2.5. The NOAEL for nose-only exposure was 0.138 mg/L and the LOAEL was 0.519 mg/L based on decreased body weight and body weight gain in males, decreased triiodothyronine and thyroxine levels in males, and inflammatory infiltrate of nasal turbinate mucosa. The NOAEL for whole-body exposure was 0.023 mg/L and the LOAEL was 0.138 mg/L based on decreased body weight and/or decreased body weight gain in males and females; pulmonary inflammation, necrosis, and granulomas; decreased triiodothyronine and thyroxine, and a non-statistically significant increase in TSH in males and females. As a result of this study, nose-only exposure was selected for the main study.

There were no compound-related deaths and clinical signs were comparable among groups in the main study. Body weights were decreased in high-dose males (-7%) with a decrease in body weight gain (-10% compared to controls). Female body weights/gains were comparable among the groups. Recovery groups had comparable body weights to controls within 1-2 weeks of cessation of treatment. Thyroxine was decreased in high-dose females at 13 weeks (-31%) as well as at 4 weeks (-25%, non-significantly) when compared to controls, but was not reduced after the 13 week recovery period. High-dose males had only slight decreases (-10 to -12%) at these 2 time periods. Triiodothyronine and thyroid stimulating hormone were unaffected by treatment. Hyperplasia of thyroid follicular epithelium was seen microscopically in 3/10 females examined in the high-dose group after 13 weeks of treatment, but not at 4 weeks or after the 13-week recovery period. Thyroid weights were comparable among the groups. Insufficient blood was available for hematological analysis of females at 4 weeks. Minor changes in hematological parameters at 13 weeks and after the 13 week recovery period were not attributed to treatment. Yellow-brown pigment in kidney cortical tubules in mid- and high-dose males and females was seen microscopically at 13 weeks, but not at 4 weeks or after the recovery period. The pigment was attributed to the elimination of a metabolite, ethylenebisthiocyanate sulfide (EBIS), and was unaccompanied by pathological changes. Residue analysis determined that mancozeb was excreted in the urine of both sexes, principally as ethylene thiourea (ETU). ETU residues were detected in the thyroid; parent compound was detected in the urine but was not analyzed for in the thyroid due to limited sample size. Analysis for EBIS was not performed.

The LOAEL in the previous review was based upon pigment found in mid-dose animals, an effect not considered toxicologically significant, and the LOAEL is now increased to the high dose. The previous review reported "respirable concentrations" whereas "analytical concentrations" are reported in this executive summary under current HED policy. The NOAEL is now 0.079 mg/L (21 mg/kg/day) and the LOAEL is 0.326 mg/L (88 mg/kg/day) based upon body weight decrements in males, thyroid hyperplasia in females, and decreased thyroxine in females. This study is classified acceptable/guideline and satisfies the guideline requirement for a subchronic inhalation study in rats.

<u>Dose and Endpoint for Risk Assessment:</u> NOAEL = 21 mg/kg/day based upon thyroid hyperplasia in females, and decreased thyroxine in females at 88 mg/kg/day (LOAEL).

Comments about Study/Endpoint: This study is appropriate because it is a route specific study that evaluated the target organ of concern.

11. Inhalation Exposure: Intermediate-Term (1- 6Months):

Study: 90-Day Inhalation Study in Rats
OPPTS 870.3465 (OPP § 82-4)

MRID No.: Accession 00159471

Executive Summary: See short-term inhalation section.

Dose and Endpoint for Risk Assessment: NOAEL = 21 mg/kg/day based upon thyroid hyperplasia in females, and decreased thyroxine in females at 88 mg/kg/day (LOAEL).

Comments about Study/Endpoint: This study is appropriate because it is a route specific study that evaluated the target organ of concern.

12. Inhalation Exposure: Long-Term (> 6 Months)

Study: 90-Day Inhalation Study in Rats
OPPTS 870.3465 (OPP § 82-4)

MRID No.: Accession 00159471

Executive Summary: See short-term inhalation section.

Dose and Endpoint for Risk Assessment: NOAEL = NOAEL = 21 mg/kg/day based upon thyroid hyperplasia in females, and decreased thyroxine in females at 88 mg/kg/day (LOAEL).

Comments about Study/Endpoint: This study is appropriate because it is a route specific study that evaluated the target organ of concern.

13. Margins of Exposure

Summary of target Margins of Exposure (MOEs) for risk assessment.

Route / Duration	Short-Term (1-30 Days)	Intermediate-Term (1 - 6 Months)	Long-Term (> 6 Months)
Occupational (Worker) Exposure			
Dermal	100	100	100
Inhalation	100	100	100
Residential (Non-Dietary) Exposure			
Oral	1000	1000	N/A
Dermal	1000	1000	1000
Inhalation	1000	1000	1000

For Occupational exposure: This is based on the conventional uncertainty factor of 100X (10X for intraspecies extrapolation and 10X for interspecies variation)

For Residential exposure: This is based on the conventional uncertainty factor of 100X (10X for intraspecies extrapolation and 10X for interspecies variation) and an additional 10X database uncertainty factor because there are datagaps for a comparative thyroid study with mancozeb and a developmental neurotoxicity study with the mancozeb metabolite, ETU).

14. Recommendation for Aggregate Exposure Risk Assessments

As per FQPA, 1996, when there are potential residential exposures to the pesticide, aggregate risk assessment must consider exposures from three major sources: oral, dermal and inhalation exposures. For short-term, intermediate-term, and long-term aggregate exposure risk assessments, the oral, dermal, and inhalation routes can be combined because a common toxicity endpoint (thyroid toxicity) was identified for the oral, dermal (oral equivalent), and inhalation routes.

III. CLASSIFICATION OF CARCINOGENIC POTENTIAL

1. Combined Chronic Toxicity/Carcinogenicity Study in Rats

Carcinogenicity Study in Rats

MRID:

Executive Summary:

In a combined chronic toxicity/carcinogenicity study (MRID 41903601), mancozeb (83.8%) was administered to Crl:CD(BR) rats (72/sex/dose) for 2 years. Initial dietary concentrations of 0, 10, 30, 65, or 375 ppm were gradually increased to the final dietary concentrations of 0, 20, 60, 125, or 750 ppm (males: 0, 0.77, 2.33, 4.38, or 30.9 mg/kg/day; females: 0, 1.06, 3.06, 6.72, or 40.2 mg/kg/day) for weeks 5-104 so that young rats would receive similar doses to mature rats. There was an interim sacrifice of 10 rats/sex/dose at 1 year with the remaining 62 rats/sex/dose sacrificed after 2 years of treatment.

There were no treatment-related effects upon mortality, clinical signs, hematology, urinalysis, or clinical chemistry, other than thyroid hormones as detailed below. Decreased body weight occurred in high-dose males (-12%) and females (-10%) at various time points compared to controls. Body weight gains for weeks 1-91 were slightly decreased in high-dose males (+256% vs 276% in controls) and females (+124% vs 145% in controls).

Relative liver weight was significantly increased in high-dose males (+29%) and absolute liver weight was increased non-significantly (+11%). Relative testes weight was increased in high-dose males at 12 months (+17%). An increased incidence and severity of bilateral retinopathy occurred in 125 ppm females and 750 ppm males and females. The incidence of unilateral retinopathy was decreased in 125 and 750 ppm females.

Thyroid toxicity in high-dose males and females included changes in thyroid hormone levels, microscopic changes, and changes in thyroid weights. Thyroxine levels were decreased beginning at 3 months and remained decreased until 18 months in males and to study termination in females. Triiodothyronine was decreased at 3 months in both sexes but rebounded at 3 months and remained elevated throughout the study, sometimes with statistical significance. Thyroid stimulating hormone was elevated beginning at 6 months until study termination, sometimes with statistical significance. Absolute and relative thyroid/parathyroid weights were increased at 24 months but not at 12 months. Microscopically, the incidence of thyroid follicular cell hypertrophy/hyperplasia was increased at 2 years, but not at 1 year, in 750 ppm males and females. The incidence of thyroid nodular follicular cell hyperplasia was also increased in 750 ppm males and females at 2 years. Thyroid follicular cell adenomas were increased in high-dose males (0/60, 1/62, 1/60, 0/58, and 20/61) and females (1/62, 1/60, 1/62, 1/61, and 6/61). Thyroid follicular cell carcinomas were increased in high-dose males (0/60, 0/62, 2/60, 2/58, 14/61) and females (0/62, 0/60, 0/62, 1/61, 4/61).

A granular yellowish-brown pigment was seen microscopically in the kidneys of male and female rats receiving 125 or 750 ppm. The pigment was unaccompanied by histopathological lesions or clinical chemistry or urinalysis abnormalities. The NOAEL/LOAEL were previously based upon this pigment, however, the pigment is not considered toxicologically significant in the absence of pathological changes and the NOAEL/LOAEL are increased from 60/125 ppm in the previous review to 125/750 ppm.

The NOAEL is now 125 ppm (male: 4.38 mg/kg/day, female: 6.72 mg/kg/day) and the LOAEL is 750 ppm (male: 30.9 mg/kg/day, female: 40.2 mg/kg/day) based upon thyroid toxicity and bilateral retinopathy.

Based upon toxicity, doses were adequate to assess carcinogenicity. Although survival in the 5 male dose groups was low (26%, 29%, 39%, 16%, and 21%), there were enough survivors to assess carcinogenicity. This study is classified acceptable/guideline and satisfies the guideline requirement for a combined chronic toxicity/carcinogenicity study in rats.

Discussion of Tumor Data

Thyroid follicular cell adenomas were increased in high-dose males (0/60, 1/62, 1/60, 0/58, and 20/61) and females (1/62, 1/60, 1/62, 1/61, and 6/61). Thyroid follicular cell carcinomas were increased in high-dose males (0/60, 0/62, 2/60, 2/58, 14/61) and females (0/62, 0/60, 0/62, 1/61, 4/61). There were statistically significant increases and significant positive trends in thyroid follicular cell adenomas, carcinomas and combined adenoma/carcinoma in both sexes of Sprague Dawley rats at doses up to 750 ppm.

Adequacy of the Dose Levels Tested

Dosing is considered adequate based upon thyroid toxicity (changes in thyroid hormone levels, microscopic changes, and changes in thyroid weights).

2. Carcinogenicity Study in Mice

MRID No.: 41981801

Executive Summary:

In a mouse carcinogenicity study (MRID 41981801), mancozeb was administered to 94 CD-1 mice/sex/dose group at dietary concentrations of 0, 30, 100, or 1000 ppm for 18 months. There was an interim sacrifice of 24 mice/sex/group at 12 months with the remaining 70 mice/sex/group sacrificed at 18 months. Doses were reported as approximately equivalent to 0, 4, 13, or 131 mg/kg/day in males and 0, 5, 18, or 180 mg/kg/day in females. However, in stability testing from weeks 52-80, much of the mancozeb had degraded to ethylenethiourea

(ETU) and an accurate conversion from ppm to mg/kg/day of mancozeb cannot be calculated. The laboratory suspected "a problem in mixing or storage of the feed occurred prior to freezing of the samples before shipment" to the testing laboratory.

Survival was not adversely affected and clinical signs were comparable among the groups. Only minor body weight decrements occurred in the high-dose group (-6% for males and -8% for females) at various time points. Body weight gain was comparable to controls at week 12. Food consumption was comparable between groups.

In comparison to other studies, minimal thyroid toxicity occurred in this study. Thyroxine was significantly reduced in high-dose males at 12 months (-56%) and in high-dose females at 12 months (-76%) and 18 months (-62%). T3 was decreased in high-dose females at 18 months (-31%) and was elevated in high-dose males at 18 months (+45%). High-dose females had an unexpected decrease in TSH at 18 months (-67%, non-significant) even though both T3 and thyroxine were decreased. There were no changes in thyroid weights or histopathology. There were slight decreases in red blood cell counts at 12 months in males (-6%) and in females (-8%) along with changes in associated hematological parameters.

The only gross pathological finding possibly due to treatment was an increase in diffuse discoloration of lymph nodes in high-dose males at termination. Relative liver weights were increased in high-dose females only at 12 months (+9%); absolute liver weights in females were increased at 12 months (+17%), but not with statistical significance. Absolute and relative liver weights in males were non-significantly reduced in comparison to controls.

The **NOAEL** for systemic toxicity is 100 ppm and the **LOAEL** is 1000 ppm based upon minor body weight decrements and changes in thyroid hormone levels. An accurate conversion from ppm to mg/kg/day of mancozeb could not be calculated due to instability of test material from weeks 52-80, when much of the mancozeb had degraded to ethylenethiourea (ETU).

There were no histopathological findings that were considered treatment-related and no treatment-related increase in tumors occurred. However, dosing was considered inadequate for assessing the carcinogenic potential due to minimal toxicity in the study (Carcinogenicity Peer Review of Mancozeb, 11/19/92).

This carcinogenicity study in mice is classified **unacceptable** and does **not** satisfy the guideline requirement for a carcinogenicity study with mancozeb in mice because dosing was inadequate for assessing carcinogenicity.

Discussion of Tumor Data

There were no treatment-related changes in tumor rates.

Adequacy of the Dose Levels Tested

There were only minor changes in body weight, thyroid hormone levels, and liver weights. Dosing was considered inadequate for assessing the carcinogenic potential due to minimal toxicity in the study (Carcinogenicity Peer Review of Mancozeb, 11/19/92).

The Cancer Assessment Review Committee did not recommend that the carcinogenicity study in mice be repeated; however, the requirement to repeat them would be reserved. Future requests for a re-evaluation of the classification of mancozeb or other EBDCs, based on a mechanistic approach to risk assessment, would warrant/necessitate submissions of an adequate carcinogenicity study in mice to assess the full carcinogenic potential for mancozeb (HED Document No. 013554, dated July 7, 1999).

3. Classification of Carcinogenic Potential

The HED Cancer Assessment Review Committee classified mancozeb as a group B2 probable human carcinogen (Carcinogenicity Peer Review of Mancozeb, 11/19/92). Since mancozeb is degraded and/or metabolized to ethylene thiourea (ETU) which causes the same types of thyroid tumors, the mancozeb carcinogenicity risk has been calculated with the Q_1^* for ETU. The Q_1^* for ETU, using a 3/4 scaling factor was determined to be 6.01 x 10^{-2} mg/kg/day^{-1} based upon female mouse liver tumors in an NTP study (memo, Bernice Fisher and Hugh Pettigrew, 2/24/95). A metabolic conversion factor of 0.075 was determined for mancozeb and other EBDC compounds for cancer risk assessments; in other words, 7.5% of mancozeb is metabolized to ETU *in vivo* (Albin Kocialski memo, 9/12/89).

The HED Cancer Assessment Review Committee met on June 9, 1999 to further evaluate the carcinogenicity of the EBDCs which are converted to ETU. The Committee concluded that mancozeb shall continue to be regulated by the Q_1^* for ETU. Based on the weight of evidence, the Committee was confident that the risk would not be underestimated with respect to cancer potential using the ETU data (HED Document No. 013554, dated July 7, 1999).

IV. MUTAGENICITY

Mancozeb has repeated negative results for both bacterial and mammalian cell gene mutation assays. Mancozeb was positive for aberrations in CHO cells, but was negative in rat bone marrow and in a mouse micronucleus assay. Mancozeb was positive in a SCE assay in CHO cells. A weak positive result was found for UDS in HeLa cells, but negative results were found in primary rat hepatocytes. In many instances the genotoxic effects of mancozeb are not substantial, as in the SCE assay, however, in some instances, the response is significantly large, as in the *in vitro* cytogenetics assay. Overall, it appears that mancozeb has some genotoxic activity that may contribute to a mutagenic concern. (Carcinogenicity Peer Review of Mancozeb, 11/19/92)

V. HAZARD CHARACTERIZATION

Mancozeb is an ethylenebisdithiocarbamate (EBDC) fungicide. The EBDC fungicides include mancozeb, metiram, and maneb, all of which share a common metabolite/degradate, ethylene thiourea (ETU).

Mancozeb is not acutely toxic *via* the oral, dermal, or inhalation routes of exposure (Tox Category IV). Mancozeb is not a skin irritant (Tox Category IV) nor is it a skin sensitizer although it did cause eye irritation (Tox Category III).

The thyroid is a target organ for mancozeb, the other EBDCs, and ETU. Thyroid toxicity in chronic and subchronic rat and dog studies with mancozeb was manifested as alterations in thyroid hormones (decreased thyroxine and increased or decreased triiodothyronine), increased thyroid weight, and microscopic thyroid lesions (mainly thyroid follicular cell hyperplasia). Thyroid stimulating hormone was also increased.

Developmental defects which occurred in the rat developmental toxicity study were numerous and severe. Although the developmental defects were of concern, they occurred at the same dose which caused maternal mortality and do not indicate increased susceptibility of offspring. Central nervous system defects (atrophy of brain tissue, cranial edema, dilated ventricles of the brain, compression and/or hemorrhages of the spinal cord, deficiency of tissue in the olfactory bulb, meningoencephalocele), skeletal system defects (incomplete cranial ossification, wide cranial sutures, curved clavicle, fused sternebrae, absent caudal or sacral vertebrae, fused and/or thickened ribs, wavy ribs, misshapen or incomplete ossification of hindlimb long bones, kyphosis, incomplete ossification or misshapen pelvis), and gross defects (agnathia, cleft palate, cleft lip, club limb, stubby tail, forelimb flexure, kinked tail, and cryptorchidism) were noted at the high dose which also caused maternal mortality. Abortions occurred in the rabbit developmental toxicity study at the high dose which also caused maternal mortality. No teratogenic effects were attributed to mancozeb in the rabbit developmental toxicity study and there was no indication of enhanced susceptibility of offspring in the rabbit developmental study.

No reproductive toxicity occurred in the 2-generation reproduction study in rats with mancozeb. Microscopic evidence of toxicity to male and female reproductive systems was noted in the subchronic dog study, but not in the chronic dog study.

An acute neurotoxicity study with mancozeb was not available. Injury to peripheral nerves (demyelination, myelin phagocytosis, Schwann cell proliferation, thickened myelin sheath, intrasheath ellipsoids, neurofibrillary degeneration, and myelin ovoids, bubbles, and debris) was seen

microscopically in the rat subchronic neuropathology study with associated clinical signs (abnormal gait and limited use of rear legs) and loss of muscle mass.

Other toxicity included increases in bilateral retinopathy in the chronic rat study. Elevated cholesterol and a mild, regenerative, anemia occurred in subchronic and chronic dog studies.

Mancozeb is rapidly absorbed and eliminated in the urine. ETU is a major metabolite in the rat, but not in the mouse. There is no evidence of bioaccumulation.

Thyroid follicular cell adenomas and carcinomas were increased in high-dose males and females in the combined rat toxicity/carcinogenicity study with mancozeb. Mancozeb is classified as a B2 human carcinogen and after applying the metabolic conversion factor for EBDC to ETU of 0.075, the Q_1* of ETU will be used (6.01×10^{-2} mg/kg/day^{-1}). Based on the weight of the evidence, it is concluded that the risk would not be underestimated with respect to cancer potential using the ETU data.

VI. ACUTE TOXICITY

Acute Toxicity Data for Mancozeb (014504)

Guideline No.	Study Type	MRID #	Results	Toxicity Category
870.1100	Acute Oral	00142522	LD_{50} > 5000 mg/kg	IV
870.1200	Acute Dermal	00142522	LD_{50} > 5000 mg/kg	IV
870.1300	Acute Inhalation	00142522	LC_{50} > 5.14 mg/L	IV
870.2400	Primary Eye Irritation	00142522	corneal damage < 7 days	III
870.2500	Primary Skin Irritation	00142522	Negative	IV
870.2600	Dermal Sensitization	40469501	Negative	N/A

VII. DATA GAPS / REQUIREMENTS

The following studies with mancozeb are required:

1) Comparative study for thyroid toxicity in adults and offspring
2) Acute Neurotoxicity
3) 21-Day dermal toxicity in rats

Although the HIARC determined previously that a developmental neurotoxicity (DNT) study was required for mancozeb (HED Doc. 0050362), this requirement will now be addressed by a DNT study with the mancozeb metabolite, ETU, because the teratogenic effects to the brain after mancozeb treatment are believed to be due to ETU. A subchronic oral study was previously required to provide a comparison of effects between oral and dermal routes of exposure, however, there are adequate subchronic studies, and an adequate dermal study can satisfy these data needs.

VIII. SUMMARY OF TOXICOLOGY ENDPOINT SELECTION

Summary of Toxicology Endpoint Selection for Mancozeb (014504)

Exposure Scenario	Dose for Risk Assessment and Uncertainty Factor	Special FQPA Safety Factor and Level of Concern	Study and Toxicological Effects
Acute Dietary Females 13-50 years age	NOAEL = 30 mg/kg/day *UF = 1000* **Acute RfD = 0.03** mg/kg/day	FQPA SF = 1X **aPAD** = acute RfD / FQPA SF = **0.03** mg/kg/day	Rabbit developmental toxicity. LOAEL = 80 mg/kg/day based on abortions.
Acute Dietary General Population including infants/children	**None**	N/A	No appropriate endpoint from oral toxicity studies.
Chronic Dietary all populations	NOAEL = 4.38 mg/kg/day *(UF = 1000)* **Chronic RfD = *0.004*** mg/kg/day	FQPA SF = 1x **cPAD** = chronic RfD / FQPA SF = **0.004** mg/kg/day	Chronic/carcinogenicity study in rats. LOAEL = 30.9 mg/kg/day based on thyroid toxicity.
Incidental Oral (Short-term)	NOAEL = 9.24 mg/kg/day	**Residential** LOC for MOE = 1000 **Occupational** = N/A	90-day dietary study in rats. LOAEL = 17.82 mg/kg/day based on decreased thyroxine.
Incidental Oral (Intermediate-term)	NOAEL = 9.24 mg/kg/day	**Residential** LOC for MOE = 1000 **Occupational** = N/A	90-day dietary study in rats. LOAEL = 17.82 mg/kg/day based on decreased thyroxine.
Dermal Short-term (1-30 days	Oral NOAEL = 9.24 mg/kg/day[1]	**Residential** LOC for MOE = 1000 **Occupational** LOC for MOE = 100	90-day dietary study in rats. LOAEL = 17.82 mg/kg/day based on decreased thyroxine.
Dermal Intermediate-term (1-6 months)	Oral NOAEL = 9.24 mg/kg/day[1]	**Residential** LOC for MOE = 1000 **Occupational** LOC for MOE = 100	90-day dietary study in rats. LOAEL = 17.82 mg/kg/day based on decreased thyroxine.
Dermal Long-term (> 6 months)	Oral NOAEL = 4.38 mg/kg/day[1]	**Residential** LOC for MOE = 1000 **Occupational** LOC for MOE = 100	Chronic/carcinogenicity study in rats. LOAEL = 30.9 mg/kg/day based on thyroid toxicity.

[1] Dermal absorption = 26%

Exposure Scenario	Dose for Risk Assessment and Uncertainty Factor	Special FQPA Safety Factor and Level of Concern	Study and Toxicological Effects
Inhalation Short-term (1-30 days)	Inhalation NOAEL = 21 mg/kg/day	**Residential** LOC for MOE = 1000 **Occupational** LOC for MOE = 100	90-day inhalation study in rats. LOAEL = 88 mg/kg/day based on thyroid hyperplasia and decreased thyroxine in females.
Inhalation Intermediate-term (1-6 months)	Inhalation NOAEL = 21 mg/kg/day	**Residential** LOC for MOE = 1000 **Occupational** LOC for MOE = 100	90-day inhalation study in rats. LOAEL = 88 mg/kg/day based on thyroid hyperplasia and decreased thyroxine in females.
Inhalation Long-term (> 6 months)	Inhalation NOAEL = 21 mg/kg/day	**Residential** LOC for MOE = 1000 **Occupational** LOC for MOE = 100	90-day inhalation study in rats. LOAEL = 88 mg/kg/day based on thyroid hyperplasia and decreased thyroxine in females.
Carcinogenicity	Q_1* for ETU = 0.0601 $(mg/kg/day)^{-1}$	B2 carcinogen with low-dose extrapolation for human risk assessment based on liver tumors in female mice with ETU.	

UF = uncertainty factor, FQPA SF = Special FQPA safety factor, NOAEL = no observed adverse effect level, LOAEL = lowest observed adverse effect level, PAD = population adjusted dose, (a = acute, c = chronic) RfD = reference dose, MOE = margin of exposure, LOC = level of concern, NA = Not Applicable

NOTE: The Special FQPA Safety Factor recommended by the HIARC **assumes** that the exposure databases (dietary food, drinking water, and residential) are complete and that the risk assessment for each potential exposure scenario includes all metabolites and/or degradates of concern and does not underestimate the potential risk for infants and children.

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Chemical:	**Mancozeb**
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